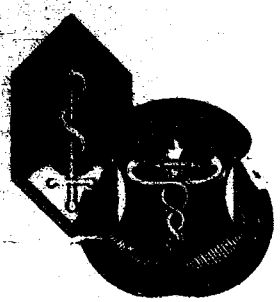

(2)

Institute Report No. 353

Acute Oral Toxicity of Nitrosoguanidine in Sprague-Dawley Rats

Earl W. Morgan, DVM, MAJ, VC
Conrad R. Wheeler, PhD
and
Don W. Korte, Jr., PhD, LTC, MSC

MAMMALIAN TOXICOLOGY BRANCH
DIVISION OF TOXICOLOGY

September 1989

Toxicology Series: 168

LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129

Acute Oral Toxicity of Nitrosoguanidine in Sprague-Dawley Rats (Toxicology Series 168)-- Morgan *et al.*

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This material has been reviewed by Letterman Army Institute of Research and there is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author(s) and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense. *(AR 360-5)*

William C. Cole 29 Sept '89

Donald G. Corby (date)
COL, MC
Commanding

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19. ABSTRACT (Continue on reverse if necessary and identify by block number)

The acute oral toxicity of nitrosoguanidine was determined in male and female Sprague-Dawley rats by using the oral gavage single-dose method. The median lethal dose for both male and female Sprague-Dawley rats was greater than 5000 mg/kg. Major clinical signs were red nasal staining, yellow perianal staining, irritability, diarrhea, ataxia, and inactivity. These signs were minimal in severity and occurred primarily during the first 24 hours after dosing. According to the classification scheme of Hodge and Sterner, these results place nitrosoguanidine in the practically non-toxic class.

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ABSTRACT

The acute oral toxicity of nitrosoguanidine was determined in male and female Sprague-Dawley rats by using the oral gavage single-dose method. The median lethal dose for both male and female Sprague-Dawley rats was greater than 5000 mg/kg. Major clinical signs were red nasal staining, yellow perianal staining, irritability, diarrhea, ataxia, and inactivity. These signs were minimal in severity and occurred primarily during the first 24 hours after dosing. According to the classification scheme of Hodge and Sterner, these results place nitrosoguanidine in the practically non-toxic class.

Key Words: Acute Oral Toxicity, Nitrosoguanidine, Munition, Nitroguanidine, Mammalian Toxicology, Rat

PREFACE

TYPE REPORT: Acute Oral Toxicity GLP Study Report

TESTING FACILITY:

US Army Medical Research and Development Command
Letterman Army Institute of Research
Presidio of San Francisco, CA 94129-6800

SPONSOR:

US Army Medical Research and Development Command
US Army Biomedical Research and Development Laboratory
Fort Detrick, MD 21701-5010
Project Officer: Gunda Reddy, PhD

PROJECT/WORK UNIT/APC: 3E162720A835/180/TLB0

GLP STUDY NUMBER: 85008

STUDY DIRECTOR: LTC Don W. Korte, Jr., PhD, MSC
Diplomate, American Board of Toxicology

PRINCIPAL INVESTIGATOR: MAJ Earl W. Morgan, DVM, VC
Diplomate, American College of
Veterinary Preventive Medicine,
American Board of Toxicology

PATHOLOGISTS: CPT Harry L. Walker, DVM, VC
MAJ Michael V. Slayter, DVM, VC

REPORT AND DATA MANAGEMENT: A copy of the final report, study protocol, SOPs, raw data, analytical, stability, and purity data of the test compound, and an aliquot of the test compound will be retained in the LAIR Archives.

TEST SUBSTANCE: Nitrosoguanidine

INCLUSIVE STUDY DATES: 13 November - 3 December 1985

OBJECTIVE: The objective of this study was to determine the acute oral toxicity of nitrosoguanidine in male and female Sprague-Dawley rats.

ACKNOWLEDGMENTS

SSG James D. Justus, BS, SP4 James J. Fischer, PFC Scott L. Schwebe, and Richard A. Spieler provided animal care.

SIGNATURES OF PRINCIPAL SCIENTISTS INVOLVED IN THE STUDY

We, the undersigned, declare that GLP Study 85008 was performed under our supervision, according to the procedures described herein, and that this report is an accurate record of the results obtained.

Don W. Korte Jr 21 Sep 89

DON W. KORTE JR., PhD / DATE
LTC, MSC
Study Director

Earl W. Morgan 24 Jun 87

EARL W. MORGAN, DVM / DATE
MAJ, VC
Principal Investigator

Conrad R Wheeler 22 Sep 89

CONRAD R. WHEELER, PhD / DATE
DAC
Analytical Chemist

DEPARTMENT OF THE ARMY

LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129-6800

REPLY TO
ATTENTION OF:

SGRD-ULZ-QA

29 September 1989

MEMORANDUM FOR Commander, LAIR

SUBJECT: QAU Clearance of Report for GLP Study 85008
Tox Series 168

I have reviewed the reply to the QAU audit conducted on 29 September 1989 and find it to be acceptable. I believe the report accurately reflects the raw data and recommend it be approved for publication.

Walter G. Bell
WALTER G. BELL
SFC, USA
Quality Assurance Officer

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Acute Oral Toxicity of Nitrosoguanidine in Sprague-Dawley Rats--Morgan *et al.*

INTRODUCTION

Nitrosoguanidine is a potential anaerobic degradation product of nitroguanidine (1), a primary component of US Army triple-base propellants, which is now produced in a Government-owned contractor-operated ammunition plant. The US Army Biomedical Research and Development Laboratory (USABRDL), as part of its mission to evaluate the environmental and health hazards of military-unique pollutants generated by US Army munitions-manufacturing facilities, conducted a review of the nitroguanidine data base and identified significant gaps in the toxicity data (2). The Division of Toxicology, LAIR, was tasked by USABRDL to develop a genetic and mammalian toxicity profile for nitroguanidine, related intermediates/by-products of its manufacture, and its environmental degradation products.

Objective of Study

The objective of this study was to determine the acute oral toxicity of nitrosoguanidine in male and female albino Sprague-Dawley rats.

MATERIALS

Test Substance

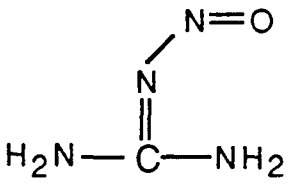
Chemical Name: Nitrosoguanidine

LAIR Code Number: TP48

Molecular Formula: CH_4N_4O

Physical State: Yellow powder

Chemical Structure:



Other chemical information is presented in Appendix A.

Vehicle

The vehicle for nitrosoguanidine was 1% gum tragacanth (Sigma Chemical Co., St. Louis, MO), expiration March 1995, in sterile water for injection (Abbott Laboratories, North Chicago, IL), expiration 1 June 1986.

Animal Data

Seven male and 7 female Sprague-Dawley rats (Bantin & Kingmar Fremont, CA) from a shipment that arrived on 12 Nov 85 were used for this study. They were identified individually with ear tags numbered 85D01144 to 85D01150 (males) and 85D01151 to 85D01157 (females) inclusive. Two males (85D01025, 85D01064) and 2 females (85D01096, 85D01121) were selected randomly from the shipment for quality control necropsy evaluation at receipt. The animal weights on 13 Nov 85 ranged from 135 to 158 g. Additional animal data appear in Appendix B.

Husbandry

Rats were caged individually in stainless-steel wire-mesh cages in racks equipped with automatically flushing dump tanks. No bedding was used in any of the cages. The diet, fed *ad libitum*, consisted of Certified Purina Rodent Chow® Diet 5002 (Ralston Purina Company, Checkerboard Square, St. Louis, MO); water was provided by continuous drip from a central line. The animal room temperature was maintained in a range from 21.1°C to 25.5°C with a relative humidity range of 36% to 53%. The photoperiod was 12 hours of light per day.

METHODS

Acclimation

The animals were acclimated for 7 days before the day of dosing. During this period they were observed daily for signs of illness.

Dose Levels

Since a pilot study indicated that the median lethal dose (MLD) of nitrosoguanidine was greater than 5000 mg/kg, a "limit dose" of 5000 mg/kg was selected for evaluating the acute oral toxicity of nitrosoguanidine.

Preparation of Compound

Nitrosoguanidine is a yellow powder that is insoluble in water or organic liquids. It was therefore suspended at a concentration of 500 mg/ml in 1% gum tragacanth. The gum tragacanth vehicle was prepared by mixing 5 g of gum tragacanth in 495 ml of sterile water using the Kinematica model CH-6010 homogenizer.

Chemical Analysis of Dosing Suspension

The nitrosoguanidine dosing suspension (500 mg/ml) was analyzed for accuracy and stability (Appendix A). The dosing suspension was shown to be 92.8% of the target concentration. Nitrosoguanidine suspensions have been shown to be stable (less than 2% decomposition) for 4 hours. To ensure the accuracy of the administered dose, the suspension was prepared immediately before administration. The entire procedure was completed within 30 minutes.

Test Procedures

This study was conducted in accordance with EPA guidelines (3) and LAIR SOP-OP-STX-36 (4).

The volume of dosing suspension each animal received was based upon the desired dose level, the concentration of the compound in the suspension, and the weight of the animal. Volumes ranged from 1.7 to 1.8 ml in the males and from 1.2 to 1.6 ml in the females. Dosing was performed using the oral gavage method without sedating the animals or administering anesthesia. Sterile disposable syringes (Becton, Dickenson & Co, Rutherford, NJ) fitted with 14 gauge, 3-inch, ball-tipped feeding tubes (Popper & Sons, Inc., New Hyde Park, NY) were used for dosing. The test

compound animals were dosed between 0756 and 0824 hours on 19 November 1985.

Observations

Observations for mortality and signs of acute toxicity were performed daily according to the following procedure: (a) animals were observed undisturbed in their cages, (b) animals were removed from their cages and given a physical examination, and (c) animals were observed after being returned to their cages. On the day of dosing, the animals were checked intermittently throughout the day. Recorded observations were performed approximately 1, 2, and 4 hours after dosing and daily for the remainder of the 2-week test period. A second "walk-through" observation was performed daily and only significant observations recorded. Body weights were recorded weekly during the course of the study.

Necropsy

All animals were submitted for a complete gross necropsy immediately after receiving a barbiturate overdose.

Duration of Study

Appendix C is a historical listing of study events.

Changes/Deviations

The study was accomplished according to the protocol and applicable amendments with the following exceptions: To conserve animals, a vehicle control group was omitted. A concurrent vehicle control group dosed with 1% gum tragacanth was used in GLP study 85015 (acute oral toxicity of JA-2 in rats) which received animals from the same shipment as this study. The feed was not removed from study animals the night before necropsy until 1822 hours. None of these changes was thought to have an effect on the study.

Storage of Raw Data and Final Report

A copy of the final report, study protocols, raw data, retired SOPs and an aliquot of the test compound will be retained in the LAIR Archives.

RESULTS

Mortality

No deaths occurred during the study.

Clinical Observations

The most frequently observed categories of clinical signs in animals administered the limit dose of nitrosoguanidine were urogenital signs (11 of 14 animals dosed), miscellaneous signs including red staining of the nose and neck and yellow staining of the perineum (10 of 14), gastrointestinal signs (6 of 14), and behavioral signs (5 of 14). The urogenital signs were predominately a very yellow coloring of the urine similiar to the color of nitrosoguanidine. A yellow color similiar to that of nitrosoguanidine was also observed in the feces of two animals although contamination of the feces with urine could not be ruled out. Gastrointestinal signs included diarrhea, bloody feces, dehydration, and emaciation. Dehydration and emaciation are often associated with difficulties with the automatic watering system. However, no obvious mechanical difficulties were reported. Behavioral signs included irritability, inactivity, ataxia, and torticollis. Most signs were observed predominately during the first 24 hours after dosing. Other signs including reddish stains on the nose and diarrhea were observed sporadically throughout the study.

Table 1 contains a summary of clinical observations. Appendix D contains individual animal histories.

Weight gains of survivors were not affected by dosing. Table 2 presents the mean body weights by groups. Appendix E contains individual weight tables.

Gross Pathology Findings

No lesions were found at necropsy that could be attributed to the test compound or the dosing procedure. The veterinary pathologist's report appears in Appendix F.

TABLE 1: Incidence Summary for Clinical Observations in Rats Administered Nitrosoguanidine (Limit Dose)

Category Clinical Signs	Group Dose (mg/kg) (N=)	1 5000 (limit) 7
MALES		
Respiratory[a]		0
Behavorial[b]		2
Gastrointestinal[c]		5
Rough Coat		0
Hunched Posture		0
Urogenital[d]		5
Miscellaneous[e]		7
FEMALES		
Respiratory[a]		1
Behavorial[b]		3
Gastrointestinal[c]		1
Rough Coat		2
Hunched Posture		1
Urogenital[d]		6
Miscellaneous[e]		3

[a] Includes panting.

[b] Includes irritability, inactivity, ataxia, and torticollis.

[c] Includes diarrhea, bloody feces, dehydration, and emaciation.

[d] Includes yellow urine and blood in urine.

[e] Includes red stains on the nose and neck, and yellow stains on the perineum.

TABLE 2: Mean Body Weights for Rats Administered Nitrosoguanidine

Group	Receipt	Dosing Day*	Day 7	Day 14*
		MALES		
5000 mg/kg	152.3 ±2.0(7)	172.0 ±1.3(7)	223.6 ±7.3(7)	235.4† ±9.7(7)
		FEMALES		
5000 mg/kg	149.7 ±3.1(7)	148.0 ±4.9(7)	183.6 ±4.3(7)	181.1† ±8.0(7)

* After an overnight fast.

† One animal was dehydrated.

DISCUSSION

The median oral lethal dose (MLD) for nitrosoguanidine is greater than 5000 mg/kg in male and female Sprague-Dawley rats. This places nitrosoguanidine in the "practically non-toxic" class (5).

The primary clinical sign was a striking yellow color of the urine similiar to the color of nitrosoguanidine. No chemical analysis of the urine was performed to confirm the presence of nitrosoguanidine. If a valid observation, it would indicate that the nitrosoguanidine was being absorbed from the gastrointestinal tract and is still practically non-toxic to the animal. However, mouse studies (6) have indicated that intraperitoneal administration of nitrosoguanidine is very toxic (lethal doses as low as 21 mg/kg) which suggests that the striking yellow color of the urine was a coincidental observation and not due to the presence of nitrosoguanidine.

CONCLUSION

Nitrosoguanidine is a practically non-toxic compound when administered orally since it produced no significant observable effects or deaths at the "limit dose" of 5000 mg/kg in male and female Sprague-Dawley rats.

REFERENCES

1. Kaplen DL, Cornell JH, Kaplen AM. Decomposition of nitroguanidine. Environ Sci Technol 1982; 16:488-492.

2. Kenyon, KF. A data base assessment of environmental fate aspects of nitroguanidine. Frederick, MD: US Army Medical Bioengineering Research and Development Laboratory, 1982; DTIC No. ADA125591.

3. Environmental Protection Agency. Office of Pesticides and Toxic Substances, Office of Toxic Substances (TS-792). Acute exposure, oral toxicity. In: Health effects test guidelines. Washington, DC: Environmental Protection Agency, August 1982; EPA 560/6-82- 0011.

4. Acute oral toxicity study (ALD and LD_{50}). LAIR Standard Operating Procedure OP-STX-36, Letterman Army Institute of Research, Presidio of San Francisco, CA. 15 June 1984.

5. Hodge HC, Sterner JH. Tabulation of toxicity classes. Amer Ind Hyg Assoc Q. 1943; 10:93-96.

6. Epstein SS, Arnold E, Andrea J, Bass W, Bishop Y. Detection of chemical mutagens by the dominant lethal assay in the mouse. Toxicol Appl Pharmacol 1972; 23:288-325.

Appendix A: CHEMICAL DATA

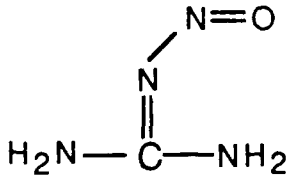
Chemical Name: Nitrosoguanidine

Chemical Abstracts Service Registry No.: 674-81-7

Lot Number: WCC-2-002

LAIR Code: TP48

Chemical Structure:



Molecular Formula: CH_4N_4O

Molecular Weight: 88

Physical State: Yellow powder

Analytical Data:

HPLC: Nitrosoguanidine was analyzed using conditions similar to those employed by Burrows *et al.*[1] Conditions were as follows: column, Brownlee RP-18 (4.6 mm x 25 cm); mobile phase, water; flowrate, 0.8 ml/min. The effluent was monitored at 255 nm. The retention times for nitrosoguanidine and nitroguanidine were 4.4 and 6 min, respectively. The HPLC data demonstrated that the nitrosoguanidine contained approximately 2.5% nitroguanidine.[2]

IR (KBr): 3378, 3096, 1690, 1649, 1508, 1341, 1266,1134, 1088, 1035, 690, 668 cm^{-1}.[3]

---

[1] Burrows EP, Brueggeman EE, Hoke SH. Chromatographic trace analysis of guanidine, substituted guanidines and striazines in water. Chromatog 1984;16:494-8.

[2] Wheeler, CR. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #84-05-010.3, p 37. Letterman Army Institute of Research, Presidio of San Francisco, CA.

[3] *Ibid.* p 30.

Appendix A (cont.): CHEMICAL DATA

Solubility:

A saturated solution of nitrosoguanidine in water was prepared at room temperature. A 1:500 dilution of this solution produced an absorbance of 0.533 units. Using an extinction coefficient of 13,305 L/moles·cm, the concentration of nitrosoguanidine in the original saturated solution was calculated to be 1.76 mg/ml.[4]

Stability:

Stable for at least 4 hours in 1% gum tragacanth at room temperature.[5]

Source: Alan Rosencrance
US Army Biomedical Research and Development Laboratory
Fort Detrick, Maryland

Analysis of Dosing Suspension:

A suspension of nitrosoguanidine (target concentration, 500 mg/ml) was prepared using 1% gum tragacanth as the vehicle and stored overnight in a refrigerator. The next day the suspension was analyzed by UV spectroscopy as follows: An aliquot of the suspension (1 ml) was transferred to a 1000 ml volumetric flask and diluted to volume with water. A portion of this solution was diluted to 50 ml in a second volumetric flask. The absorbency of the final dilution was determined at 255 nm. The following data were used to calculate the concentration of nitrosoguanidine in the dosing suspension: 1.376 AVFS; extinction coefficient at 255 nm, 13,050; all path length, 1 cm; dilution factor, 50,000. The concentration of the dosing suspension was calculated to be 464 mg/ml or 92.8% of the target value.[6]

---

[4] Wheeler CR. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #85-01-006, p 66. Letterman Army Institute of Research, Presidio of San Francisco, CA.

[5] Wheeler, CR. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #85-12-022, p 11-13. Letterman Army Institute of Research, Presidio of San Francisco, CA.

[6] Wheeler, CR. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #84-05-010.3, p 72-74. Letterman Army Institute of Research, Presidio of San Francisco, CA.

Appendix B: ANIMAL DATA

Species: *Rattus norvegicus*

Strain: Sprague-Dawley

Source: Bantin & Kingman, Fremont, CA

Sex: Male and female.

Dates of birth: Male: 2 October 1985
Female: 25 September 1985

Animals in each group: 7 males and 7 females

Condition of animals at start of study: Normal

Body weight range at dosing: 121-175 g

Identification procedures: Ear tag: tag numbers 85D01144 to 85D01150 (males) and 85D01151 to 85D01157 (females), inclusive.

Pretest conditioning: Quarantine/acclimation 12 - 18 November 1985

Justification: The laboratory rat has proven to be a sensitive and reliable system for lethal dose determination.

Appendix C: HISTORICAL LISTING OF STUDY EVENTS

Date	Event
12 Nov 85	Received shipment of Sprague-Dawley rats. Rats were checked for physical condition, sexed, and individually caged.
13 Nov 85	Animals were weighed and ear-tagged. Two male and 2 female rats were submitted to necropsy for quality control.
13-18 Nov 85	Animals were observed daily.
18 Nov 85	Rats were weighed and food was removed from the animals by 1800.
19 Nov 85	Animals were weighed, dosed, and observed at 1, 2, and 4 hours after dosing.
20 Nov-2 Dec 85	All animals were observed daily in a.m. and p.m.
26 Nov 85	All animals weighed.
2 Dec 85	Food removed by 1822 hours.
3 Dec 85	Animals were weighed, observed for clinical signs, and submitted for necropsy.

Appendix D: INDIVIDUAL ANIMAL HISTORIES

MALE: 5000 mg/kg NITROSOGUANIDINE

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85D01144	Stain, Nose, Red	Nov. 19,Dec.2,3	Slight
	Inactive	Nov. 19	Slight
	Urine, Yellow	Nov. 19	Slight
85D01145	Stain, Nose, Red	Nov. 19,Dec.2	Slight
	Diarrhea	Dec. 3	Present
85D01146	Stain, Nose, Red	Nov. 19	Slight
	Urine, Yellow	Nov. 19	Slight
	Blood in Feces	Nov. 19	Slight
85D01147	Torticollis	Nov. 19	Slight
	Inactive	Nov. 19	Slight
	Diarrhea	Nov. 19,Dec.3	Slight
	Stain, Perianal, Yellow	Nov. 19	Slight
85D01148	Stain, Nose, Red	Nov. 19	Slight
	Urine, Yellow	Nov. 19	Slight
	Diarrhea	Nov. 22	Slight
	Dehydrated	Dec. 2,3	Marked
85D01149	Diarrhea	Nov. 19	Slight
	Urine, Yellow	Nov. 19	Slight
	Stain, Nose, Red	Nov. 19	Slight
	Stain, Perianal, Yellow	Nov. 21	Slight
85D01150	Stain, Nose, Red	Nov. 19	Slight
	Urine, Yellow	Nov. 19	Slight
	Stain, Neck, Red	Nov. 20	Slight

Appendix D (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 5000 mg/kg NITROSOGUANIDINE

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85D01151	Rough Coat	Nov. 19	Slight
	Stain, Perianal, Yellow	Nov. 19	Slight
	Blood in Urine	Nov. 20	Moderate
	Panting	Nov. 20	Moderate
85D01152	Irritable	Nov. 20	Slight
	Urine, Yellow	Nov. 20	Slight
85D01153	Stain, Nose, Red	Nov. 19	Slight
	Urine, Yellow	Nov. 19,20	Slight
	Irritable	Nov. 20	Slight
85D01154	Urine, Yellow	Nov. 19	Slight
	Diarrhea	Nov. 22	Slight
85D01155	Hunched Posture	Nov. 19	Moderate
	Stain, Perianal, Yellow	Nov. 19	Slight
	Inactive	Nov. 19	Slight
	Ataxia	Nov. 20	Slight
	Rough Coat	Nov. 20	Slight
	Dehydrated	Dec. 2	Moderate
	Emaciated	Dec. 2,3	Moderate
85D01156	Urine, Yellow	Nov. 19	Slight
	Feces, Yellow	Nov. 20	Slight
85D01157	Urine, Yellow	Nov. 19	Slight
	Feces, Yellow	Nov. 20	Slight

Appendix E: INDIVIDUAL BODY WEIGHTS (grams)

5000 mg/kg

Animal Number	At Receipt (g)	At Dosing (g)*	Day 7 (g)	Termination Day 14 (g)*
		MALES		
85D01144	155	175	236	277
85D01145	145	165	200	218
85D01146	158	174	196	221
85D01147	156	175	234	244
85D01148	145	171	244	200†
85D01149	151	172	237	254
85D01150	156	172	218	234
Mean	152.3	172.0	223.6	235.4
Standard Deviation	5.4	3.5	19.2	25.5
Standard Error of the Mean	2.0	1.3	7.3	9.7
		FEMALES		
85D01151	144	145	176	177
85D01152	157	157	199	195
85D01153	158	159	190	196
85D01154	148	150	188	194
85D01155	135	121	164	137†
85D01156	153	156	189	192
85D01157	153	148	179	177
Mean	149.7	148.0	183.6	181.1
Standard Deviation	8.1	13.2	11.5	21.1
Standard Error of the Mean	3.1	4.92	4.3	8.0

* Animals weighed after an overnight fast.
† Animal dehydrated.

Appendix F: PATHOLOGY REPORT

GLP Study #85008

Principal Investigator: MAJ Morgan APC#: LLEO

I. INTRODUCTION

Study: Nitrocellulose-Nitroguanidine
Animal: Rat/Sprague-Dawley/male and female
Reference: SOP-OP-STX-36.

II. HISTORY

Acute Oral Toxicity (MLD). Limit test (5000 mg/kg). No significant clinical signs were noted.

III. SUMMARY OF PROCEDURES

Euthanasia: Sodium Pentobarbital.
Fixative: 10% buffered formalin.
Histopathology: None.
Clinical Lab: None.

IV. GROSS FINDINGS

DOSE GROUP 1 - MALE

LAIR ACC#	ANIMAL ID#	OBSERVATION
38609	85D01144	Not Remarkable (NR)
38610	85D01145	NR
38611	85D01146	NR
38612	85D01147	NR
38613	85D01148	NR
38614	85D01149	NR
38615	85D01150	NR

DOSE GROUP 1 - FEMALE

38616	85D01151	NR
38617	85D01152	NR
38618	85D01153	NR
38619	85D01154	Lung - conjestion, diffuse, mild
38620	85D01155	NR
38621	85D01156	NR
38622	85D01157	NR

Appendix F (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85008

V. GROSS SUMMARY:

All tissues examined were within normal limits except for animal 38619 for which the original LAIR Form 54 was misplaced and no gross comment can be made on this animal.

VI. HISTOPATHOLOGY OBSERVATIONS:

LAIR 38619: Lung - congestion, diffuse, mild.

VII. HISTOPATHOLOGY SUMMARY:

Mild pulmonary congestion was an isolated incidental finding and was probably unrelated to the test compound.

VIII. SUMMARY COMMENTS:

The isolated finding of pulmonary congestion is considered incidental. Other tissues examined were within normal limits.

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15 October 1986

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